



SUBSTITUTE SEQUENCE LISTING

<1> Rosenberg, Stephen P.

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Rebar, Edward J.

Sangamo Biosciences, Inc.

<120> Selection of Sites for Targeting by Zinc Finger
Proteins and Methods of Designing Zinc Finger Proteins
to Bind to Preselected Sites

<130> 019496-001810US

<140> US 09/825,242

<141> 2001-04-02

<160> 97

<170> PatentIn Ver. 2.1

<210> 1

<211> 25

<212> PRT

<213> Artificial Sequence

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<223> Description of Artificial Sequence:exemplary motif
characterizing the C-2H-2 class of zinc finger
proteins (ZFP)

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<222> (1)..(25)

<223> Xaa = any amino acid

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<222> (4)..(5)

<223> Xaa = any amino acid, may be present or absent

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<223> Xaa = any amino acid, may be present or absent

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1

5

10

15

Xaa Xaa His Xaa Xaa Xaa Xaa His

20

25

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<400> 2
Thr Gly Glu Lys Pro
1 5

<210> 3
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Gly Gly Gly Gly Ser
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Leu Arg Gln Lys Asp Gly Gly Gly Ser Glu Arg Pro
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<211> 16

<212> PRT

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<213> Mus sp.

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<223> DNA binding domain of mouse transcription factor
 Zif268

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Tyr Ala Cys Pro Val Glu Ser Cys Asp Arg Arg Phe Ser Arg Ser Asp
 1 5 10 15

Glu Leu Thr Arg His Ile Arg Ile His Thr Gly Gln Lys Pro Phe Gln
 20 25 30

Cys Arg Ile Cys Met Arg Asn Phe Ser Arg Ser Asp His Leu Thr Thr
 35 40 45

His Ile Arg Thr His Thr Gly Glu Lys Pro Phe Ala Cys Asp Ile Cys
 50 55 60

Gly Arg Lys Phe Ala Arg Ser Asp Glu Arg Lys Arg His Thr Lys Ile
 65 70 75 80

His Leu Arg Gln Lys
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 531-624 in Sp-1 transcription factor

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Pro Gly Lys Lys Lys Gln His Ile Cys His Ile Gln Gly Cys Gly Lys
 1 5 10 15

Val Tyr Gly Lys Thr Ser His Leu Arg Ala His Leu Arg Trp His Thr
 20 25 30

Gly Glu Arg Pro Phe Met Cys Thr Trp Ser Tyr Cys Gly Lys Arg Phe
 35 40 45

Thr Arg Ser Asp Glu Leu Gln Arg His Lys Arg Thr His Thr Gly Glu
 50 55 60

Lys Lys Phe Ala Cys Pro Glu Cys Pro Lys Arg Phe Met Arg Ser Asp
 65 70 75 80

His Leu Ser Lys His Ile Lys Thr His Gln Asn Lys Lys Gly
 85 90

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 transcription factor consensus sequence

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Met Glu Lys Leu Arg Asn Gly Ser Gly Asp Pro Gly Lys Lys Lys Gln
 1 5 10 15

His Ala Cys Pro Glu Cys Gly Lys Ser Phe Ser Lys Ser Ser His Leu
 20 25 30

Arg Ala His Gln Arg Thr His Thr Gly Glu Arg Pro Tyr Lys Cys Pro
 35 40 45

Glu Cys Gly Lys Ser Phe Ser Arg Ser Asp Glu Leu Gln Arg His Gln
 50 55 60

Arg Thr His Thr Gly Glu Lys Pro Tyr Lys Cys Pro Glu Cys Gly Lys
 65 70 75 80

Ser Phe Ser Arg Ser Asp His Leu Ser Lys His Gln Arg Thr His Gln
 85 90 95

Asn Lys

<210> 11

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binding site

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containing three D-able subsites

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with two overlapping D-able subsites

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<400> 15
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<210> 18

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<400> 21
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<400> 22
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<400> 28
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motif searched by protocol 1

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24

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motif searched by protocol 1

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motif searched by protocol 2

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<221> modified_base

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<210> 37

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<212> DNA

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motif searched by protocol 2

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<221> modified_base

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motif searched by protocol 2

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<210> 39
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motif searched by protocol 2

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<210> 40
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<210> 42
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<210> 43
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<210> 44
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<210> 45
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motif searched by protocol 2

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motif searched by protocol 2

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motif searched by protocol 2

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<222> (12)..(13)

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<212> DNA

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motif searched by protocol 2

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<400> 50
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motif searched by protocol 2

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<400> 51
knnknnknngg nnnknnknngg nnn

23

<210> 52
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motif searched by protocol 2

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<400> 52
knnknnknngg nnknnknnkn gg

22

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motif searched by protocol 2

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<210> 54
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motif searched by protocol 2

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knnknnknngg nnknngnnn

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<211> 19

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence:target site DNA
motif searched by protocol 2

<220>

<221> modified_base

<222> (1)..(19)

<223> n = g, a, c or t

<400> 56

knnknnknngg nnknnknngg

19

<210> 57

<211> 22

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence:target site DNA
motif searched by protocol 3

<220>

<221> modified_base

<222> (1)..(22)

<223> n = g, a, c or t

<220>

<221> modified_base

<222> (10)..(12)

<223> n = g, a, c or t, may be present or absent

<400> 57

kngknnknnn nnknngknnkn nn

22

<210> 58

<211> 23

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence:target site DNA
motif searched by protocol 3

<220>

<221> modified_base
<222> (1)..(23)
<223> n = g, a, c or t

<220>
<221> modified_base
<222> (11)..(13)
<223> n = g, a, c or t, may be present or absent

<400> 58
kngknnknnn nnnkngknnk nnn

23

<210> 59
<211> 22
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence:target site DNA
motif searched by protocol 3

<220>
<221> modified_base
<222> (1)..(22)
<223> n = g, a, c or t

<220>
<221> modified_base
<222> (10)..(12)
<223> n = g, a, c or t, may be present or absent

<400> 59
kngknnknnn nnknnkngkn nn

22

<210> 60
<211> 23
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence:target site DNA
motif searched by protocol 3

<220>
<221> modified_base
<222> (1)..(23)
<223> n = g, a, c or t

<220>
<221> modified_base
<222> (11)..(13)
<223> n = g, a, c or t, may be present or absent

<400> 60
kngknnknnn nnnknnkngk nnn

23

<210> 61
<211> 22
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence:target site DNA
motif searched by protocol 3

<220>
<221> modified_base
<222> (1)..(22)
<223> n = g, a, c or t

<220>
<221> modified_base
<222> (10)..(12)
<223> n = g, a, c or t, may be present or absent

<400> 61
kngknnknnn nnknnknnkn gk.

22

<210> 62
<211> 23
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence:target site DNA
motif searched by protocol 3

<220>
<221> modified_base
<222> (1)..(23)
<223> n = g, a, c or t

<220>
<221> modified_base
<222> (11)..(13)
<223> n = g, a, c or t, may be present or absent

<400> 62
kngknnknnn nnnknnknnk ngk

23

<210> 63
<211> 22
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence:target site DNA
motif searched by protocol 3

<220>
<221> modified_base
<222> (1)..(22)
<223> n = g, a, c or t

<220>
<221> modified_base
<222> (10)..(12)
<223> n = g, a, c or t, may be present or absent

<400> 63
knnkngknnn nnkngknnkn nn

22

<210> 64
<211> 23
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence:target site DNA
motif searched by protocol 3

<220>
<221> modified_base
<222> (1)..(23)
<223> n = g, a, c or t

<220>
<221> modified_base
<222> (11)..(13)
<223> n = g, a, c or t, may be present or absent

<400> 64
knnkngknnn nnnkngknnk nnn

23

<210> 65
<211> 22
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence:target site DNA
motif searched by protocol 3

<220>
<221> modified_base
<222> (1)..(22)
<223> n = g, a, c or t

<220>
<221> modified_base
<222> (10)..(12)
<223> n = g, a, c or t, may be present or absent

<400> 65

knnkngknnn nnknnkngkn nn

22

<210> 66
<211> 23
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence:target site DNA
motif searched by protocol 3

<220>
<221> modified_base
<222> (1)..(23)
<223> n = g, a, c or t

<220>
<221> modified_base
<222> (11)..(13)
<223> n = g, a, c or t, may be present or absent

<400> 66
knnkngknnn nnnknnkngk nnn

23

<210> 67
<211> 22
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence:target site DNA
motif searched by protocol 3

<220>
<221> modified_base
<222> (1)..(22)
<223> n = g, a, c or t

<220>
<221> modified_base
<222> (10)..(12)
<223> n = g, a, c or t, may be present or absent

<400> 67
knnkngknnn nnknnknnkn gk

22

<210> 68
<211> 23
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence:target site DNA
motif searched by protocol 3

<220>
 <221> modified_base
 <222> (1)..(23)
 <223> n = g, a, c or t

<220>
 <221> modified_base
 <222> (11)..(13)
 <223> n = g, a, c or t, may be present or absent

<400> 68
 knnkngknnn nnnknnknnk ngk

23

<210> 69
 <211> 22
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> Description of Artificial Sequence:target site DNA
 motif searched by protocol 3

<220>
 <221> modified_base
 <222> (1)..(22)
 <223> n = g, a, c or t

<220>
 <221> modified_base
 <222> (11)..(12)
 <223> n = g, a, c or t, may be present or absent

<400> 69
 knnknnkngk nnkngknnkn nn

22

<210> 70
 <211> 23
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> Description of Artificial Sequence:target site DNA
 motif searched by protocol 3

<220>
 <221> modified_base
 <222> (1)..(23)
 <223> n = g, a, c or t

<220>
 <221> modified_base
 <222> (12)..(13)
 <223> n = g, a, c or t, may be present or absent

<400> 70
knnknnkngk nnnkngknnk nnn

23

<210> 71
<211> 22
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence:target site DNA
motif searched by protocol 3

<220>
<221> modified_base
<222> (1)..(22)
<223> n = g, a, c or t

<220>
<221> modified_base
<222> (11)..(12)
<223> n = g, a, c or t, may be present or absent

<400> 71
knnknnkngk nnknnkngkn nn

22

<210> 72
<211> 23
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence:target site DNA
motif searched by protocol 3

<220>
<221> modified_base
<222> (1)..(23)
<223> n = g, a, c or t

<220>
<221> modified_base
<222> (12)..(13)
<223> n = g, a, c or t, may be present or absent

<400> 72
knnknnkngk nnnknnkngk nnn

23

<210> 73
<211> 22
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence:target site DNA

motif searched by protocol 3

<220>

<221> modified_base

<222> (1)..(22)

<223> n = g, a, c or t

<220>

<221> modified_base

<222> (11)..(12)

<223> n = g, a, c or t, may be present or absent

<400> 73

knnknnkngk nnknnknnkn gk

22

<210> 74

<211> 23

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence:target site DNA
motif searched by protocol 3

<220>

<221> modified_base

<222> (1)..(23)

<223> n = g, a, c or t

<220>

<221> modified_base

<222> (12)..(13)

<223> n = g, a, c or t, may be present or absent

<400> 74

knnknnkngk nnnknnknnk ngk

23

<210> 75

<211> 19

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence:target site DNA
motif searched by protocol 3

<220>

<221> modified_base

<222> (1)..(19)

<223> n = g, a, c or t

<400> 75

knnknnkngk ngknnknnn

19

<210> 76
<211> 19
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence:target site DNA
motif searched by protocol 3

<220>
<221> modified_base
<222> (1)..(19)
<223> n = g, a, c or t

<400> 76
knnknnkngk nnkngknnn 19

<210> 77
<211> 19
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence:target site DNA
motif searched by protocol 3

<220>
<221> modified_base
<222> (1)..(19)
<223> n = g, a, c or t

<400> 77
knnknnkngk nnknnkngk 19

<210> 78
<211> 10
<212> DNA
<213> Glycine max

<220>
<223> soybean FAD2-1 cDNA ZFP target segment FAD 1

<400> 78
gaggtagagg 10

<210> 79
<211> 10
<212> DNA
<213> Glycine max

<220>
<223> soybean FAD2-1 cDNA target segment FAD 2

<400> 79

gtcgtgtgga

10

<210> 80

<211> 10

<212> DNA

<213> Glycine max

<220>

<223> soybean FAD2-1 cDNA target segment FAD 3

<400> 80

gttgaggaag

10

<210> 81

<211> 10

<212> DNA

<213> Glycine max

<220>

<223> soybean FAD2-1 cDNA target segment FAD 4

<400> 81

gaggtggaag

10

<210> 82

<211> 10

<212> DNA

<213> Glycine max

<220>

<223> soybean FAD2-1 cDNA target segment FAD 5

<400> 82

taggtggtga

10

<210> 83

<211> 12

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence:test sequence

<400> 83

agtgcgcggt gc

12

<210> 84

<211> 10

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence:target site
with base immediately to the 3' side of target
site

<400> 84
agtgcgcggt

10

<210> 85
<211> 10
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence:target site
with base immediately to the 3' side of target
site

<400> 85
gtgcgcggtg

10

<210> 86
<211> 10
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence:target site
with base immediately to the 3' side of target
site

<400> 86
tgcgcggtgc

10

<210> 87
<211> 10
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence:target site
with base immediately to the 3' side of target
site

<220>
<221> modified_base
<222> (10)
<223> n = undefined

<400> 87
gcgcggtgcn

10

<210> 88
<211> 7

<212> PRT
<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: finger F3 for
ordered output from optimal design target site

<400> 88

Glu Arg Asp His Leu Arg Thr
1 5

<210> 89

<211> 7

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: finger F2 for
ordered output from optimal design target site

<400> 89

Arg Ser Asp Glu Leu Gln Arg
1 5

<210> 90

<211> 7

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: finger F1 for
ordered output from optimal design target site

<400> 90

Arg Lys Asp Ser Leu Val Arg
1 5

<210> 91

<211> 7

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: finger for
disordered output from optimal design target site

<400> 91

Arg Ser Asp Glu Leu Thr Arg
1 5

<210> 92

<211> 7

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: finger for
disordered output from optimal design target site

<400> 92

Arg Ser Asp Glu Arg Lys Arg
1 5

<210> 93

<211> 21

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: three finger
ZFP design using F3, F2 and F1 fingers for ordered
output from optimal design target site

<400> 93

Arg Lys Asp Ser Leu Val Arg Arg Ser Asp Glu Leu Gln Arg Glu Arg
1 5 10 15

Asp His Leu Arg Thr
20

<210> 94

<211> 21

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: ZFP sequence
(F1, F2 and F3) from SBS design GR-223

<400> 94

Arg Ser Ala Asp Leu Thr Arg Arg Ser Asp His Leu Thr Arg Glu Arg
1 5 10 15

Asp His Leu Arg Thr
20

<210> 95

<211> 21

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: ZFP sequence
(F1, F2 and F3) from Zif 268

<400> 95

Arg Ser Asp Glu Leu Thr Arg Arg Ser Asp His Leu Thr Thr Arg Ser

